

### **AMENDMENTS TO THE CLAIMS**

Prior to examination please amend the claims in PCT/AU2005/000262 as indicated below. The following listing of the claims replaces all prior listings of the claims:

1. [original] A host-cell free method for culturing *Cryptosporidium* comprising the step of introducing *Cryptosporidium*, at a first lifecycle stage, into a host-cell free medium under conditions which enable the *Cryptosporidium* to progress to a second lifecycle stage.
2. [original] A method according to claim 1 wherein the first and second lifecycle stages are selected from the group consisting of: oocyst including excysted oocysts, sporozoite, trophozoite, meront 1, merozoites (Type 1), meront 11 (early), meront 11 (late), merozoites (type 11), macrogamont, microgamete and zygote.
3. [original] A method according to claim 1 wherein the first lifecycle stage is an oocyst or a sporozoite and the second lifecycle stage is an oocyst, sporozoite or a trophozoite.
4. [original] A method according to claim 1 wherein the second lifecycle stage is an oocyst.
5. [original] A host-cell free method for culturing *Cryptosporidium* comprising the step of introducing *Cryptosporidium*, at a first lifecycle stage, into a host-cell free medium under conditions which enable the *Cryptosporidium* to complete its lifecycle.
6. [original] A host-cell free method for producing *Cryptosporidium* biomass from an initial inoculum of *Cryptosporidium* comprising the steps of: (i) putting the inoculum into a host cell free medium; and (ii) culturing the *Cryptosporidium* to increase the *Cryptosporidium* biomass.
7. [currently amended] A method according to ~~any one of the preceding claims~~ claim 1 wherein the host cell free medium is a buffered and balanced combination of inorganic salts, amino acids and vitamins.
8. [original] A method according to claim 7 wherein the medium further comprises an additional constituent selected from the group consisting of: a carbohydrate source, antibiotics, bile and serum.

9. [currently amended] A method according to ~~any one of the preceding claims~~  
claim 1 wherein the medium has a pH at or about neutral pH.

10. [currently amended] A method according to ~~any one of the preceding claims~~  
claim 1 wherein the host cell free medium further comprises a second phase in the form of  
serum that has been treated to render it viscous or semi-solid.

11. [original] A method according to claim 10 wherein the serum is coagulated.

12. [currently amended] A method according to claim 10 ~~or 11~~ wherein the  
serum used to form the second phase is foetal calf serum.

13. [original] A host-cell free method for culturing *Cryptosporidium* comprising the  
steps of: a. isolating *Cryptosporidium* oocysts; b. excysting the isolated oocysts; c.  
resuspending the excysted oocysts in a host-cell free culture medium; d. incubating the  
culture prepared in step (c) under suitable conditions; and e. harvesting oocysts from the  
medium.

14. [currently amended] A method according to ~~any one of the preceding claims~~  
claim 1, wherein the *Cryptosporidium* belongs to the species selected from the group  
consisting of: *Cryptosporidium anderson*, *Cryptosporidium parvum*, *Cryptosporidium muris*,  
*Cryptosporidium hominis*, *Cryptosporidium wrairi*, *Cryptosporidium felis*, *Cryptosporidium*  
*canis*, *Cryptosporidium baileyi*, *Cryptosporidium meleagridis*, *Cryptosporidium galli*,  
*Cryptosporidium serpentis*, *Cryptosporidium saurophilum* and *Cryptosporidium molnari*.

15. [original] A host cell free medium capable of maintaining *Cryptosporidium* or  
enabling the progress of *Cryptosporidium* through its lifecycle, the medium comprising a  
buffered and balanced combination of inorganic salts, amino acids, vitamins and additional  
constituents.

16. [original] A biphasic host cell free medium capable of maintaining  
*Cryptosporidium* or enabling the progress of *Cryptosporidium* through its lifecycle the  
medium comprising a buffered and balanced combination of inorganic salts, amino acids,  
vitamins and additional constituents.

17. [currently amended] A medium according to claim 15 ~~or 16~~ wherein the additional constituents are selected from the group consisting of: amino acid supplements, carbohydrate source, antibiotics, bile and serum.

18. [currently amended] A medium according to ~~any of claims 15 to 17~~ claim 15 with a pH about neutral.

19. [original] A medium according to claim 16 wherein the second phase comprises serum that has been treated to render it viscous or semi-solid.

20. [original] A medium according to claim 19 wherein the serum is foetal calf serum.

21. [original] A method for preparing an immunogenic preparation comprising at least one *Cryptosporidium* antigen, the method comprising the steps of: (i) introducing *Cryptosporidium*, at a first lifecycle stage, into a host-cell free medium under conditions which enable the *Cryptosporidium* to progress to a second lifecycle stage; (ii) isolating the *Cryptosporidium* at the second lifecycle stage; and (iii) preparing a therapeutic preparation using the *Cryptosporidium* isolated from step (ii).

22. [original] A method according to claim 21 wherein the second lifecycle stage is anextracellular lifecycle stage.

23. [original] A method according to claim 21 wherein the second lifecycle sage is a trophozoite, merozoite or otherextracellular gamont-like stage.

24. [currently amended] A therapeutic composition comprising a therapeutically effective amount of *Cryptosporidium* cultured according to ~~any one of claims 1 to 14~~ claim 1 and a physiologically acceptable carrier.

25. [original] A composition according to claim 24 comprising a whole cell extract of one or more *Cryptosporidium* lifecycle stages.

26. [original] A composition according to claim 25 comprising one or more *Cryptosporidium* lifecycle stages that have been treated to disrupt their cellular structure.

27. [original] A composition according to claim 24 comprising at least one isolated and purified *Cryptosporidium* antigen.

28. [original] A composition according to claim 26 wherein the cellular disruption has been achieved by a technique selected from the group consisting of: sonication, osmotic pressure, freezing, exposure to detergents such as sodium dodecyl sulfate (SDS), and heating.

29. [currently amended] A composition according to ~~any one of claims 24 to 28~~ claim 24 wherein the *Cryptosporidium* cells have been inactivated.

30. [currently amended] A method of preventing or treating a disease associated with *Cryptosporidium* infection in a subject comprising administering to the subject a therapeutically effective amount of a composition according to ~~any one of claims~~ claim 24 to 29.

31. [original] A method for detecting *Cryptosporidium* in a sample comprising the steps of: (i) subjecting the sample to the culture method described herein; and (ii) detecting the *Cryptosporidium*.

32. [original] A method for detecting *Cryptosporidium* in a sample comprising the steps of: (i) introducing the sample into a host-cell free medium under conditions which enable *Cryptosporidium* to progress to a further lifecycle stage; and (ii) detecting the *Cryptosporidium*.

33. [original] A method for detecting *Cryptosporidium* in a sample comprising the steps of (i) introducing the sample into a host-cell free medium under conditions which enable the *Cryptosporidium* to complete its lifecycle ; and (ii) detecting the *Cryptosporidium*.

34. [currently amended] A method according to ~~any one of claims 31 to 33~~ claim 31 wherein the sample is from a water source that is to be used by humans or animals.

35. [original] A method according to claim 34 wherein the water source is a source of drinking water such as a dam, lake, river or rain catchment area.

36. [currently amended] A method according to ~~any one of claims 31 to 35~~ claim 31 wherein the *Cryptosporidium* is detected via visual examination.

37. [original] A method according to claim 36 wherein the visual examination is via a microscope or some other means that enables any *Cryptosporidium* in the sample to be viewed.

38. [currently amended] A method according to ~~any one of claims 31 to 35~~ claim 31 wherein the *Cryptosporidium* is detected using PCR.

39. [currently amended] A method according to ~~any one of claims 31 to 38~~ claim 31 further comprising the step of pretreating the sample to concentrate any *Cryptosporidium* therein.

40. [original] A method according to claim 39 wherein the pre-treatment comprises centrifugation of the sample.